Received Date : 26-Jun-2016

Revised Date : 22-Sep-2016

Accepted Date : 23-Sep-2016

Article type : Standard Papers

Editor: Nate Sanders

Section heading: Community Ecology

Decoupled diversity dynamics in green and brown webs during primary succession in a salt marsh

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SUMMARY

1. Terrestrial ecosystems are characterised by a strong functional connection between the green (plant-herbivore-based) and brown (detritus-detritivore-based) parts of the food web, which both develop over successional time. However, the interlinked changes in green and brown food web diversity patterns in relation to key ecosystem processes are rarely studied.
2. Here, we demonstrate changes in species richness, diversity and evenness over a wide range of invertebrate green and brown trophic groups during 100 years of primary succession in a salt marsh ecosystem, using a well-calibrated chronosequence.
3. We contrast two hypotheses on the relationship between green and brown food web diversity across succession: i) 'coupled diversity hypothesis', which predicts that all trophic groups covary similarly with the main drivers of successional ecosystem assembly versus ii) the 'decoupled diversity hypothesis', where green and brown trophic groups diversity respond to different drivers during succession.
4. We found that, while species richness for plants and invertebrate herbivores (green web groups) both peaked at intermediate productivity and successional age, the diversity of

macro-detritivores, microarthropod microbivores and secondary consumers (brown web groups) continuously increased towards the latest successional stages. These results suggest that green web trophic groups are mainly driven by vegetation parameters, such as the amount of bare soil, vegetation biomass production, and vegetation height, while brown web trophic groups are mostly driven by the production and standing stock of dead organic material and soil development.

5. Our results show that plant diversity cannot simply be used as a proxy for the diversity of all other species groups that drive ecosystem functioning, as brown and green diversity components in our ecosystem responded differently to successional gradients.

KEY WORDS: brown web, chronosequence, ecosystem assembly rules, ecosystem functioning, food web, green web, salt marsh, succession

INTRODUCTION

Succession is one of the oldest recognized processes in ecology (Clements 1916; Elton 1958). Since the earliest days of successional research, many studies have investigated how diversity changes along succession, but with a strong emphasis on aboveground taxa, especially plants (Clements 1916; Miles & Walton 1993). Compositional changes in connected trophic levels (herbivores, predators, decomposers) have long been (implicitly) assumed to be the derivative of such plant-driven succession (Connell & Slatyer 1977). We therefore lack an understanding of how other trophic groups, especially those below ground, change across successional axes. This is unfortunate, because the current biodiversity crisis urges us to develop basic understanding of

how diversity changes along environmental gradients such as those driven by succession. This calls for the study of the joint changes in diversity of different trophic groups, both above (the green web, i.e. plant-herbivores-based) and below ground (the brown web, i.e. detritus-detritivore-based) along successional gradients (Schrama, Olff & Berg 2012; Mulder *et al.* 2013; Crutsinger *et al.* 2014).

Two main hypotheses can be formulated on the general relationship between diversity change in trophic green and brown groups and succession. i): the "Coupled diversity hypothesis" and the ii) "Uncoupled diversity hypothesis". Diversity of the green and brown web trophic groups can be coupled if they covary simultaneously with the main drivers of ecosystem assembly (e.g. with increases in plant biomass during succession) and/or respond to each other´s diversity. In this case we would expect unimodal patterns for all trophic groups, as plant diversity usually peaks at intermediate levels of succession of plant biomass (Grime 1977; Rosenzweig 1995; Fraser *et al.* 2015). One of the main explanations for this pattern is the intermediate disturbance hypothesis (Grime 1973), which predicts that plant diversity is highest in intermediate successional stages as these are also the locations with intermediate levels of disturbance, where both early successional disturbance-tolerating and late successional disturbance intolerant species can occur. This hypothesis might be particularly important if plant diversity drives consumer diversity of both green and brown food web compartments (bottom-up regulation of diversity), or *vice versa* (*sensu* Hooper *et al.*, 2000). For example, this would occur when more plant diversity leads to more resource diversity for herbivores and decomposers (Hutchinson 1959) and in turn to more diversity in their enemies (Srivastava *et al.* 2009). Experimental evidence of this has been found (Brown & Southwood 1983; Edwards-Jones & Brown 1993; Knops *et al.* 1999; Siemann,

Haarstad & Tilman 1999). Examples of animal diversity driving plant diversity have been shown by Allan & Crawley (2011) and Hooper (2000). In contrast, the uncoupled diversity hypothesis might be best in describing diversity patterns when different ecosystem properties or processes or environmental conditions determine the diversity of each group of organisms independently. So while the coupled diversity hypothesis assumes that trophic interactions between below and above ground webs are important in explaining diversity patterns, the uncoupled diversity hypothesis assumes a larger role for abiotic conditions, which have different effects on above and below ground organisms. Evidence for the uncoupled diversity hypothesis is found in observational studies which have studied other environmental gradients than succession. For example, several studies have shown that grassland biodiversity can be a poor predictor of different groups of arthropod diversity (Kruess & Tscharntke 2002; van Klink *et al.* 2014; Manning *et al.* 2015). In addition, studies have reported that herbivore diversity depends not so much on plant diversity, but rather on plant productivity and vegetation structure (Olff & Ritchie 1998; Mittelbach *et al.* 2001; Hawkins & Porter 2003). Also for secondary consumers such as spiders, below ground invertebrates and birds, vertical vegetation structure has been argued as the main driver of their species richness (Greenstone 1984; Langellotto & Denno 2004). Although earlier work (Schrama, Berg & Olff, 2012) has shown that the biomass of both above and below ground organisms increase throughout succession in this study system, biodiversity may or may not be linked with biomass (Adler *et al*., 2011) .Therefore we insufficiently understand whether diversity of above and below ground organisms increase similarly over succession, or whether the patterns are different.

We tested these ideas in a salt marsh primary succession chronosequence, in which the diversity dynamics of dominant green and brown trophic groups was studied to the species level. This

chronosequence has been calibrated to a period of 100 years (Olff *et al.* 1997; Schrama, Olff & Berg 2012). Primary succession leads to (directional) build-up of plant biomass and corresponding changes in plant species diversity (Clements 1916; Tilman 1982; Olff *et al.* 1997; Olsen *et al.* 2011; Schrama, Olff & Berg 2012), as well as to changes in invertebrate and vertebrate species composition (Edwards & Sugg 1993; Chapin *et al.* 1994; Kaufmann 2001). This particular successional sequence is highly suitable for testing our ideas as 1) the different stages are spatially close together (within 1-3 km); 2) the rather low species richness makes the study logistically feasible; and 3) most plant species, and to a lesser extent invertebrates, are dispersed by water, as the area frequently floods with seawater. Therefore, we can exclude dispersal limitation as a possible cause of changes in ecosystem structure, making the interpretation of observed patterns in species richness easier. It therefore provides an ideal setting to study species richness patterns for different trophic groups in response to biotic and abiotic drivers, and in response to each other.

METHODS

Description of salt marsh chronosequence

The species diversity of the different brown and green trophic groups were studied on the salt marsh of the barrier island of Schiermonnikoog (53°30' N, 6°10' E), the Netherlands, in July 2008. The average yearly temperature on the island is 10.2 °C (± SD 0.72 °C), average annual rainfall is 824 mm (± SD 149.1 mm) (data from the Royal Netherlands Meteorological Institute KNMI at www.knmi.nl). On this salt marsh a well-described chronosequence is present, which spans one hundred years of succession (Figure 1; Olff *et al.* 1997; Schrama, Olff & Berg 2012).

The earliest stages are formed on the east side of the island, late succession stages are situated 8 km to the west. The justification of the space-for-time replacement of this succession has been validated by monitoring permanent vegetation plots and key soil parameters in different successional stages during the last 20 years (Olff *et al.* 1997; Schrama, Olff & Berg 2012). Along this gradient of successional stages, vegetation composition changes from a state with very sparse vegetation (dominated by *Salicornia europaea* and *Puccinellia maritima)* towards a species-rich vegetation in intermediate succession stages (dominated by *Festuca rubra*, *Limonium vulgare* and *Plantago maritima*) and finally a tall, species-poor vegetation (dominated by *Elytrigia atherica*) in late succession stages.

Sampling design

We focused on diversity patterns at seven successional stages, which were estimated as 0, 10, 25, 35, 45, 55 and 100 years of succession age in 2008 (Electronic Appendix, Figure S1). More details on vegetation composition in the different successional stages can be found in Electronic Appendix, Table S1. Salt-marsh age at each successional stage was estimated from topographic maps, aerial photographs, and the thickness of the sediment layer accumulated on top of the underlying, fixed sand layer (Olff *et al.* 1997; Schrama, Olff & Berg 2012). The sites were selected to have an equal base elevation (position at the initial elevation gradient on the bare sand flats with a base elevation of 1.16 m (± SE 2.2 cm above Amsterdam Ordnance Level (NAP). Different base elevations will have different inundation regimes, and therefore each elevation has its characteristic vegetation successional stage (Olff *et al.* 1997). As the salt marsh matures, sediment trapping by the vegetation increases the elevation of the soil surface with 16 cm of clay over 100 years of salt marsh succession, reducing the inundation frequency by floods at spring

tide (Schrama, Olff & Berg 2012). In each of the seven successional stages, five sampling points were selected with a minimum distance of 50 m between points.

To assess diversity of the above and below ground food web, we employed five different collection techniques at each of the 35 sampling points. Vascular plant species diversity, including the cover of bare soil was estimated in one 2x2 m quadrat using the cover estimation method (Londo 1976). One enclosed pitfall trap (Ø 10 cm, placed in one corner of an 50x50 cm Perspex enclosure, adapted after Lang (2000)) filled with ethylene glycol was used to gain quantitative estimates on the diversity of soil inhabiting macro-invertebrates, such as landhoppers, spiders, beetles and bugs. One closed insect-emergence trap of 50x50 cm, 80 cm height (Fig. 1) of which collection bottle was filled with ethanol (70%) were used to collect flying insects emerging from the soil and vegetation (Ausden 2000). These two methods were used in the period 4-22 July and were emptied every third day for a period of 18 days in order to completely 'empty' the plot. Accumulation curves for common species indeed showed a strong leveling off towards the end of this period, suggesting that the time of sampling was sufficient to catch most specimens (Electronic Appendix, Figure S2). Soil meso- and larger microfauna (e.g., springtails, oribatid mites, and enchytraeid worms) was extracted from a single soil core taken with an metal auger (Ø10 cm, 5 cm height) using a Tullgren funnel extraction method (Van Straalen & Rijninks 1982).

Environmental parameters

To understand how environmental variables underlie relationships between diversity and succession, we sampled the following environmental parameters: living and dead plant biomass, vegetation height, soil temperature, cover of bare soil, clay layer thickness, soil anoxia and soil moisture. We sampled above ground plant biomass at a randomly assigned location within each

sampling point. Standing biomass was clipped in one 50x50 cm squares, up to 1 cm above the soil. These samples were sorted by hand into dead and living biomass, and weighed to the nearest 0.1 g after being dried (70°C, 48 h). Vegetation height was determined at three randomly assigned sites, two meters apart, at each of the sampling points, using a 20 gr drop-disc (Ø20 cm). Soil temperature was measured continuously (to the nearest 0.5° Celsius) during the measurement period, using one iButtons (Maxim Corp.) per sampling point. An average of five measurements of the clay layer thickness (± 1mm) was taken using a clay ogre at each sampling point. Given that soil anoxia is an important driver of species distributions on salt marshes (Davy *et al.* 2011), we measured the soil redox potential at each of the 35 sampling points once in the Autumn of 2009. For this we used four measuring Platinum-electrodes (Pt) and one AgCl - reference electrode (Cole-Palmer®), which were all connected to a Graphtec GL200 Datalogger (Graphtec GB Ltd). Pt-electrodes were placed in a square design, 10 cm from the reference-electrode, at a depth of 2cm in the soil. To standardize the measurements, we read out redox potential values 2 minutes after the electrodes were placed. Averages of the four Pt-electrodes were corrected for the value of the AgCl-reference electrode (+197 mV at pH7). Although this measurement does not provide absolute values of the amount of oxygen in situ, it gives a reasonable estimate for the relative differences in anoxia between locations measured on the same day (van Bochove, Beauchemin & Theriault 2002). Soil moisture was measured using a ThetaProbe® moisture meter twice during the measuring period, respectively 7 and 14 days after the traps were put in the field.

Division of species into trophic groups

Invertebrates were identified up to the species level and afterwards merged into trophic groups according to the classification in Electronic Appendix, Table S2, using feeding preferences for each species of invertebrate according to Remmert (1983), Irmler & Heydemann (1986), Berg *et al.* (2004), Krantz & Walter (2009) and Caballero *et al.* (2004). We distinguished six trophic groups. 1. Herbivores (sap suckers and leaf chewers: weevils (Coleoptera: Curculionidae), plant- and leafhoppers (Hemiptera: Auchenorrhyncha), aphids (Hemiptera: Aphididae), leaf beetles (Coleoptera: Chrysomelidae) and moths (Lepidoptera)); 2. Algae consuming species (mainly snails (Mollusca) and rove beetles (Coleoptera: Staphylinidae)); 3. Microbivores (small soil fauna <4mm) feeding on litter colonizing microorganisms, such as springtails (Collembola) and oribatid mites (Acari: Oribatida); 4. Macro-detritivores (larger fauna feeding on litter or litter colonizing microorganisms, such as Diptera-larvae, the landhopper *Orchestia gammarellus*, and snails); 5. Below ground carnivores (predatory mites (Acari: Mesostigmata, Prostigmata) and other below ground predatory species, feeding preferably on microbivores); 6. Above ground carnivores (spiders (Araneae), ground beetles (Coleoptera: Carabidae), ladybird beetles (Coleoptera: Coccinellidae) and plant bugs (Hemiptera: Miridae)).

Statistical analysis

Relationships between the biodiversity (richness, diversity or evenness) values of each of the six different trophic groups, the summed richness of all groups together ('total richness'), the summed richness of green web trophic groups ('green web richness'), the summed richness of brown web trophic groups ('brown web richness') and the environmental parameters with salt marsh age were described using four different, competing general linear models. These were (i) a

null-model, i.e. a model without a salt marsh age term, (ii) a model with only a linear salt marsh age term, (iii) a model with both a linear and quadratic salt marsh age term. We finally selected the model with the lowest AIC, thus correcting for the number of terms included in models (Electronic Appendix, Table S3).

Statistics were carried out in R (R 2011), Sigmaplot 12.5 and Statistica 9.0.

To examine whether richness levels of different trophic groups responded differently to successional stages and other, succession-related environmental factors, we (i) conducted, a nonmetric multidimensional scaling (NMDS) analysis (a non-parametric multivariate analysis) and (ii) calculated correlations of the first two ordination axes with the measured environmental variables and the measured richness axes. Hence, the NMDS analysis, a nonmetric alternative to PCA, yielded insights in the extent to which sampling locations differed in richness patterns within and between successional stages. This analysis, which was based on the seven different species richness variables within each trophic group, was used to determine the Bray-Curtis distances among the diversity variables between the 35 sampling points, for which the metaMDS function was used (R vegan package; Oksanen, 2015; R Core Team, 2014).

After NMDS analysis, we also investigated the relationships of the richness of each trophic group and the environmental variables with the first two ordination axes, in order to investigate whether (i) richness of different trophic groups varied in their relationships with successional stage and (ii) whether different trophic groups varied in their relationships with other environmental factors.

RESULTS

Diversity dynamics of green and brown webs

We found a clear optimum for total richness at intermediate succession (Fig 2A). However, we observed strong differences between patterns of species richness for the green and brown webs. Species richness in the green web showed a clear optimum at intermediate succession (Fig 2B), whereas the brown web species richness showed a steady increase towards late succession (Fig 2C). In contrast, the Shannon diversity of either the green or brown web did not show any change over succession (Fig. 2D-F). Also the species evenness did not show any clear trends (Fig. 2G-I).

Diversity dynamics of specific trophic groups

For vascular plants, species richness first increased with successional age, but towards late succession exhibited a strong decline in the number of species (Fig. 3). Towards late succession, the species composition becomes dominated by *Elytrigia atherica*.

Within the green web, we found different species richness patterns for algae consumers, invertebrate herbivores and above ground predators. Species richness for algae consumers decreased with successional stage (Fig. 3C), whereas species richness for invertebrate herbivores showed a peak at intermediate succession stages (Fig. 3B). Both trophic groups had a very low number of species in the latest successional stage. Species richness for above ground predators showed peak in intermediate successional stages, thus mirroring the pattern of invertebrate herbivores (Fig. 3A).

Species richness of trophic groups in the brown web showed a strikingly different pattern. In general, species richness of brown web trophic groups exhibited a linear, continuous increase towards later successional stages. Especially microbivores showed a strong increase in species richness along the succession gradient (Fig. 3E). Also macro-detritivores showed an increase in species richness, which leveled off towards late succession (Fig. 3F). Below ground predators, consisting mostly of predatory mites, also showed a steady increase in species richness along succession, with a sudden increase after 45 years (Fig. 3G).

Overall, few trophic groups showed strong patterns for Pielou's evenness and Shannon diversity (H'). Significant correlations were found only for Shannon diversity of plants and below ground carnivores, which showed a clear optimum at intermediate succession (Figs 3D & 3G). Macro-detritivore diversity and evenness showed a marked decrease towards late succession. This strong trend can be ascribed to the overwhelming dominance of *Orchestia gammarellus* in later stages of succession.

Changes in important environmental factors

We found a clear saturating decrease over succession in the amount of bare soil (Fig. 4A). Furthermore, we found an increase in the depth of the sediment layer over succession (Fig. 4B). Vegetation height started below 1 cm in the first stage of succession and increases up to 26 cm towards the last successional stage (Fig. 4C). Vegetation biomass exhibited a clear increase over succession, but leveled off towards the older stages (Fig 4D), whereas litter layer depth and standing dead biomass did not show a leveling off (Fig 4E-F). Soil moisture decreased over succession (Fig. 4G) but we found a much lower value for the earliest stage of succession where the clay layer was absent. Soils were oxic in early succession (Fig 4H) and became more anoxic

as the depth of clay layer increased. The 45-year successional stage was most anoxic (Fig 4H), but soils became rapidly oxic in the later stages.

Relationship between food web composition and environmental factors

The NMDS analysis based on species richness per trophic group resulted in clear diversity differences in the subsequent stages of succession, as centroids for the species richness of the trophic groups clearly aggregated with different stages of succession (Fig. 5A). NMDS axis 1 was negatively correlated with the percentage of bare soil ($r = -0.85$; $p<0.001$) and positively with soil moisture ($r = 0.77$; $P < 0.001$). NMDS axis 2 was strongly and negatively correlated with soil redox potential ($r = -0.79$, $P < 0.001$) and although significant, less strongly with litter accumulation ($r = -0.54$, $p<0.001$) and thickness of the clay layer ($r = -0.47$, $P < 0.001$). Richness of algal consumers aggregated with the earliest stage of succession, whereas plant species richness and herbivore species richness aggregated with intermediate succession stages. Macro-detritivore species richness, microbivore species richness and above and below ground predator species richness aggregated with the later stages of succession (Fig 5A). The stress-value for this analysis (0.133) indicated a reliable outcome (Oksanen *et al.* 2011; R Core Team, 2014).

NMDS analyses based on Pielou's evenness and Shannon diversity explained a much smaller amount of variation in in food web compositional differences between the different successional stages (Figs 5 B & 5C) and correlations with the environmental factors were also much weaker. Also stress values for these analyses indicated a less reliable outcome, 0.215 and 0.200 for the analysis based on Shannon-diversity and species evenness, respectively.

DISCUSSION

Along the investigated gradient in primary succession, our results show strikingly different patterns for changes in species richness in the green and brown compartments of the food web. Trophic groups in the green web showed an increase from early successional stage followed by a decrease in species richness towards later stages of succession, whereas trophic groups in the brown web continued to increase in richness towards the later successional stages. Our results therefore support the hypothesis that brown web richness becomes decoupled from green web species richness along the studied chronosequence.

Our NMDS analysis indicated that this discrepancy in green and brown web species richness patterns along the chronosequence is likely caused by different environmental factors driving their diversity. While above ground predictors (vegetation diversity and structure) were strongly related to changes in green web species richness, belowground predictors (soil variables and dead organic matter accumulation) were much stronger related to brown web species richness.

A potential problem with studies on community assembly along chronosequences is that current conditions at early successional stages may not necessarily reflect past conditions at older locations (Pickett 1989; Mattews 1992). However, the development of vegetation patterns along the chronosequence on Schiermonnikoog has been tested extensively in long-term studies, showing that the different ages indeed reflect different temporal stages in vegetation succession (Olff *et al.* 1997; Schrama, Olff & Berg 2012). Also, the lack of large topographic variation on this salt marsh prevents variation in climatic conditions associated with small differences in elevation, which is often the case in chronosequences on glaciers and volcanic substrates

(Kaufmann 2001). Species colonisations from adjacent areas are often a confounding factor in diversity measurements on chronosequences. However, our above ground (closed traps) and below ground (soil cores from which specimens were extracted in the lab) sampling methods were specifically designed to minimize such effects.

The time scales along which food web assembly operates along our studied chronosequence were relatively short (100 years), much faster than for most other studies on successional community assembly (Mattews 1992; Kaufmann 2001; Hodkinson, Coulson & Webb 2004; Laliberté, Zemunik & Turner 2014), which helps in the assumption of space-for time substitution. This fast succession reflects the high rates of clay accretion in combination with external nutrient input in our study system by regular flooding with slightly eutrophicated seawater. Rates of ecological succession may be even higher in mainland salt marshes (Bakker *et al.* 2002), where clay accretion often happens at a much faster rate. Nonetheless, many of the measured environmental variables show similar and directional trends to those in other studies on chronosequences. For example, the amount of bare soil and soil moisture clearly decreased over the chronosequence whereas soil organic matter build-up, standing live biomass and vegetation height increased along succession. These findings correspond with many other chronosequence studies (del Moral 1993; Kaufmann 2001; Hodkinson, Coulson & Webb 2004), and suggest that the observed changes in species diversity patterns along our chronosequence likely represent similar changes in other primary succession sequences.

We find the highest diversity of species in the green web in intermediate successional stages where we also found the highest plant species diversity and highest vegetation structural

complexity. Both factors have been shown to be important for explaining herbivore species richness (Brown & Southwood 1983; Brown & Ewel 1987; Steffan-Dewenter & Tscharntke 1997; Siemann, Haarstad & Tilman 1999; Hawkins & Porter 2003; Rickert *et al.* 2012). Interestingly, most studies that report a positive effect of plant species richness on above ground arthropod richness, were done at experimental sites with artificial plant communities (Knops *et al.* 1999; Siemann, Tilman & Haarstad 1999; Symstad, Siemann & Haarstad 2000), whereas studies that report a strong influence of vegetation structure on herbivore arthropod richness were mostly done in natural communities, where vegetation structure and plant species richness often show a hump-shaped relationship with productivity (Hawkins & Porter 2003; Rickert *et al.* 2012; van Klink *et al.* 2014). Therefore, we suggest that both plant species richness and structural complexity (both highest at intermediate productivity and late successional age) explain invertebrate herbivore species richness in green webs.

Our results also showed that brown web species richness increases continuously during succession, without a drop and later successional stages. Hence, this increase in brown web species richness coincides with a decrease in plant species diversity, an increase in dead plant biomass and an increase in the depth of the oxygenated layer, which is in agreement with a number of previous studies (Scheu & Schulz 1996; Carlson *et al.* 2010). Especially for microbivore richness and belowground predator species richness, we found a major and continuous increase in species richness over succession. We suggest that the relationship between brown web diversity and dead plant biomass may have been caused by several mechanisms. First, this pattern could have been caused by a time-lag effect, where diversity in the brown web could react much slower to changes in plant species diversity than green web

diversity. While theoretically this mechanism may explain the observed effects, we deem it not very likely as we did not observe the slightest decrease in brown species richness even towards the latest successional stage. Second, we observed the development of a thick litter layer towards later stages of succession. This may be the result of a decrease in vegetation quality and a coinciding decrease of (vertebrate) herbivory (Van de Koppel *et al.* 1996; Olff *et al.* 1997; Van der Wal *et al.* 2000). Later stages of salt marsh succession are often dominated by couch grass *E. atherica*, which is of low quality to both vertebrate herbivores (Kuijper, Nijhoff & Bakker 2004) and detritivores (Hemminga & Buth 1991). If this hypothesis is correct, we would predict that a less dominant green web in late succession may lead to lower leaf consumption and a corresponding increase in litter C:N-ratio, followed by an increase in dead organic matter production. Indeed, biomass of green web trophic groups has been previously shown to decrease strongly along succession (Schrama et al. 2012). In addition, accumulation of low quality litter, with a high C:N-ratio and a low degradation rate (Berg & Staaf 1980; McClaugherty *et al.* 1985), may cause a further build-up of a litter layer. The increase in depth of the litter layer may in turn result in an increase in the amount of niches (Berg & Bengtsson 2007; Krab *et al.* 2010; Berg 2012). In addition to the development of a thick litter layer, below ground bioturbation activity may result in an increase in niche space. In our study system, an increase in below ground niche variation could be related to activity of the burrowing amphipod *Orchestia gammarellus*. This species has its highest abundances in late successional mat-forming grasses (Schrama et al. 2015). We suggest that this burrowing behaviour may lead to a well-aerated soil with a deeper vertical stratification of soil factors (Moore & Francis 1985; Schrama *et al.* 2013), hence creating opportunities for more species to co-occur (Berg 2012, Raynaud *et al.*, 2013). This is in line with other studies that find that vertical stratification of soils is a strong predictor of

community structure of soil-dwelling invertebrates (Berg & Bengtsson 2007; Kaspari & Yanoviak 2009). Our results thus suggest that a larger resource supply may be especially important for generalist detritivores. Other studies have found that many detritivores are indeed generalist (Krab, *et al.*, 2013), in the sense that they are not related to a single plant species. This may explain why the overall diversity of brown web organisms increases even in late successional stages, despite decreasing plant species diversity. However, more research into the mechanisms underlying these patterns is needed to yield conclusive evidence for this interpretation. Hence, the simultaneous result of the increase in depth of the litter layer and the oxygenated soil provide explanations for the observed continuing increase in brown web diversity over succession.

There are several indications that the observed patterns in species richness are linked to changes in ecosystem functioning. For example, our results indicate that the increase in species richness in the brown web towards later successional stages coincides with an increase in nutrient cycling (Van Wijnen & Bakker, 1999), much in line with theoretical predictions on the relationship between decomposer diversity and rate of nutrient cycling (Loreau 2001). These changes in nutrient recycling are associated with late-successional, taller plant species, which in turn repell herbivores (Kuijper, Nijhoff & Bakker 2004). Indeed, we observed a strong decrease in richness in the green web towards later successional stages, suggesting that changes in the brown and green web may be functionally linked. However, we would like to stress that, in order to show the causality of these links between diversity and function, it is necessary to carry out diversity experiments, studies on functional genes and/or studies that link observed changes in richness to the microbial community composition.

In summary, our study highlights that diversity in different parts of the food web exhibit contrasting responses to environmental gradients, thus emphasizing the need for a joined study of the dynamics of both above ground, plant-herbivore-based (green web) and below ground, detritus-detritivore-based (brown web) species richness simultaneously. Different trophic groups can highly vary in their impact on ecosystem processes and function (Duffy *et al.* 2007; Lefcheck *et al.* 2015), thus stressing the potential importance of our results for the ecosystem functions and services that human well-being depends on.

Acknowledgements

We thank Vereniging Natuurmonumenten for granting us permission for the field work on the salt marsh of Schiermonnikoog. We also thank all colleagues and friends who helped during the collection of the data. We are grateful to Paul Beuk, Theodoor Heijerman, Berend Aukema, Oscar Vorst, Ping-Ping Chen and Maurice Janssen for assistance on the identification of the specimens that belong to green web groups. Roel van Klink is gratefully acknowledged for help on the NMDS. MS, HO and MPB designed the research; MS carried out the field work; MS analyzed the data together with FvdP; MS, FvdP, HO and MPB wrote the paper. The authors declare they have no conflict of interest regarding this manuscript. This work was funded through a NWO PIONIER grant to HO.

Data Accessibility

Data are available from the Dryad Digital Repository: http://dx.doi.org/10.5061/dryad.s636m (Schrama et al. 2016).

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Figure captions

Figure 1. 100-year chronosequence at the salt marsh of the barrier island of Schiermonnikoog, The Netherlands. Black dots indicate sampling locations at every stage of vegetation succession in this study. At every stage of succession, measurements were done on species diversity of green and brown web trophic groups and environmental variables. The pictures above the map give an impression of the vegetation structure. Sample site of two years of succession was not used in this study. Grey shadings indicate the gradual increase of the island to the east over the last 200 years, which was derived from topographic maps and aerial pictures (for further details, see Olff *et al*., 1997).

Figure 2. Changes in overall species richness, diversity and evenness along the successional gradient. From left to right, the panels give the total food web, green web and brown web composition, respectively. From top to bottom, the panels show patterns for species richness (A-C), Shannon diversity (H') (D-F) and Pielou's evenness (E) G-I). Fits were based on AIC values after model selection. Only those fits that include a linear or quadratic term are shown. Fits were calculated using all five sampling points per location, with R^2 and *P*-value at the bottom of each of the graphs.

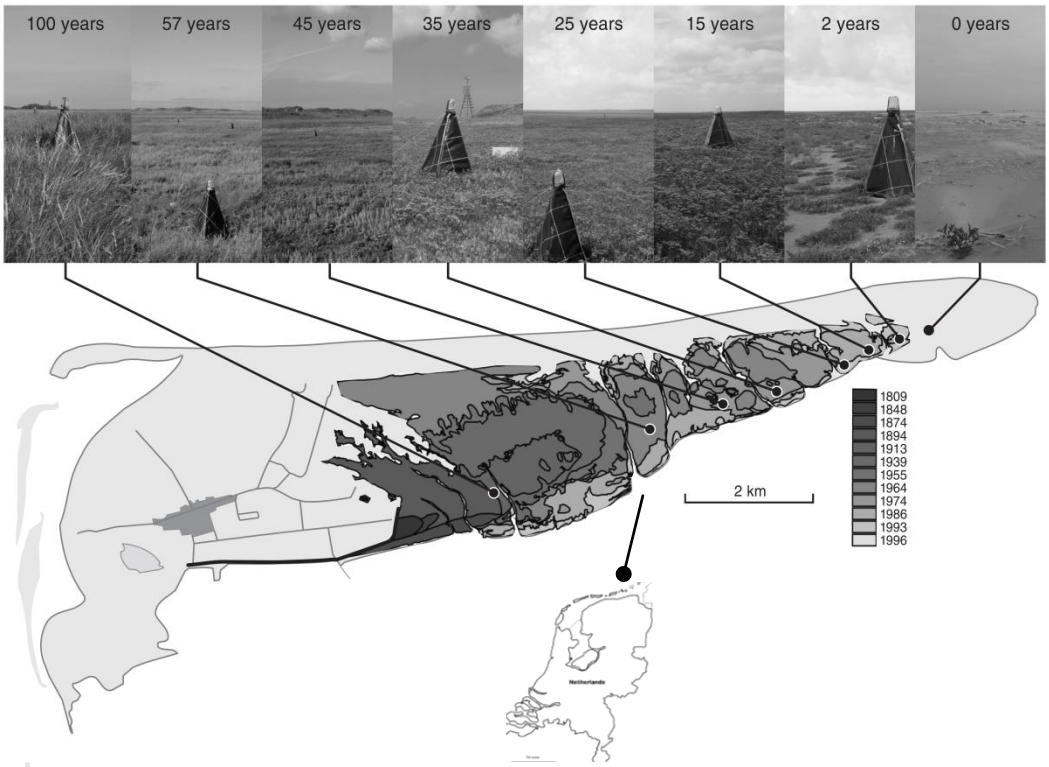
Figure 3. Diversity for all trophic groups along the succession gradient. From left to right, the panels show patterns for species richness, Shannon diversity (H') and Pielou's evenness (E). Each row (A-G) gives results for a different trophic group. Open symbols indicate green web trophic groups (± SE); closed symbols indicate brown web trophic groups (± SE). Fits were based on AIC values after model selection. Only those fits that include a linear or quadratic term are shown. Fits were calculated using all five sampling points per location, with R^2 and *P*-value at the bottom of each of the graphs.

Figure 4. Changes in environmental and abiotic parameters over the chronosequence. Bars represent standard errors. A) The percentage of bare soil (± SE) decreased with successional age. B) Sediment layer depth (cm) ± SE increased with succession to about 16 cm thick. C) Vegetation height (cm) increased strongly towards late succession. D) Standing live biomass increased, but levels off towards late succession. E) Litter layer increased linearly towards late succession. F) Standing live biomass increased linearly towards late. G) Soil moisture as a percentage of soil volume (± SE) first showed a strong increase and a gradual decreasing trend towards late succession. H) Soil redox potential at 2 cm depth decreased strongly in the first stages of succession and then increased again. Panels A, B, C and G were previously published in Schrama et al. (2012). Fits were based on AIC values after model selection. Only those fits that include a linear or quadratic term are shown. Fits were calculated using all five sampling points per location, with R^2 and *P*-value at the bottom of each of the graphs.

Figure 5. NMDS plots of trophic group diversity along the succession gradient. A) NMDS based on species richness in each of the trophic groups, B) NMDS based on species evenness (Pielou's E) in each of the trophic groups, and C) NMDS based on the Shannon diversity index (H'). Age of the different succession stages is shown in given in each polygon and is indicated with a

different color. Vectors indicate different environmental variables and are scaled proportionally in each of the graphs. Closed grey circles depict centroids for each of the trophic groups: above ground pred: Above ground predators.

Figure 1.



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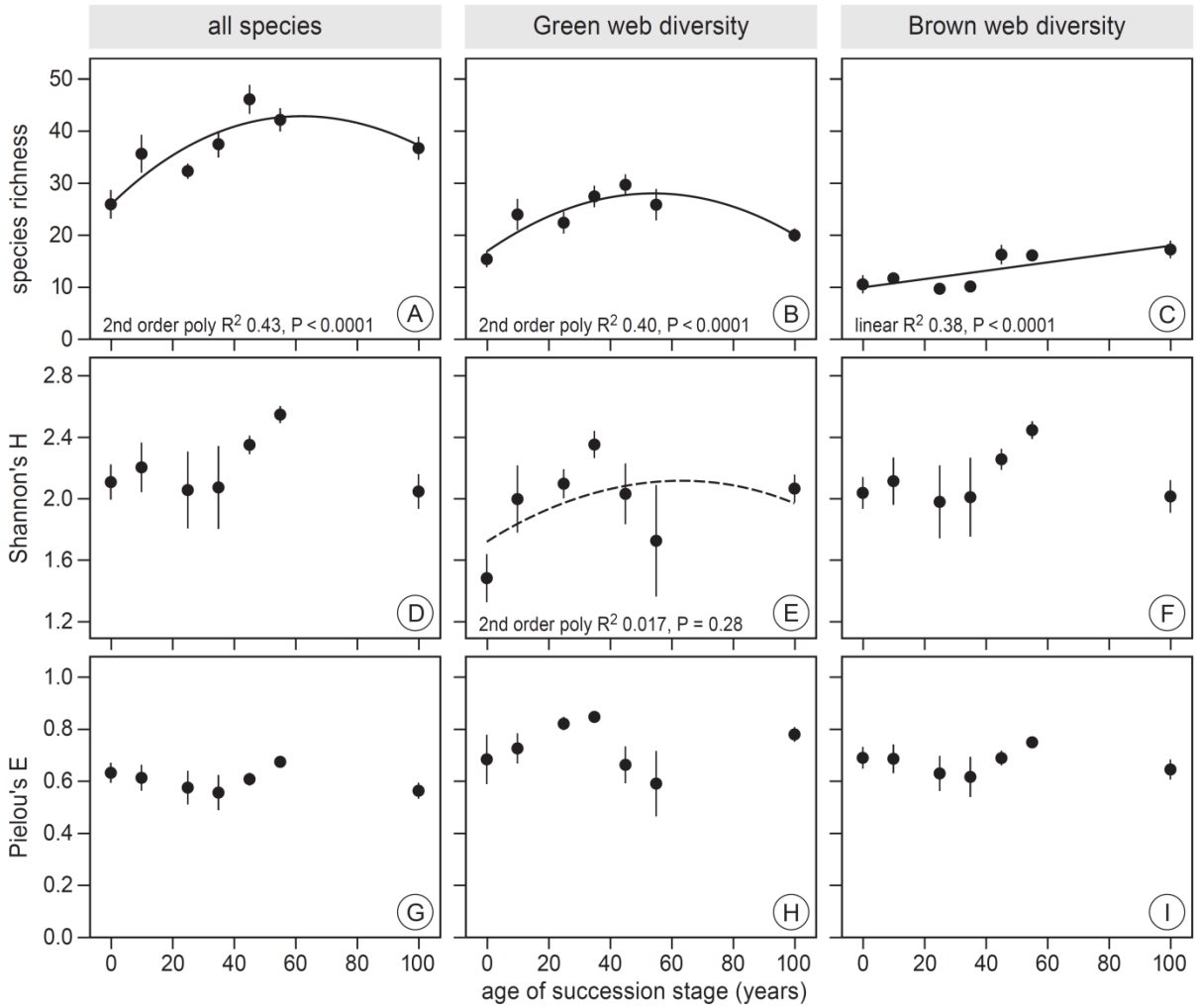

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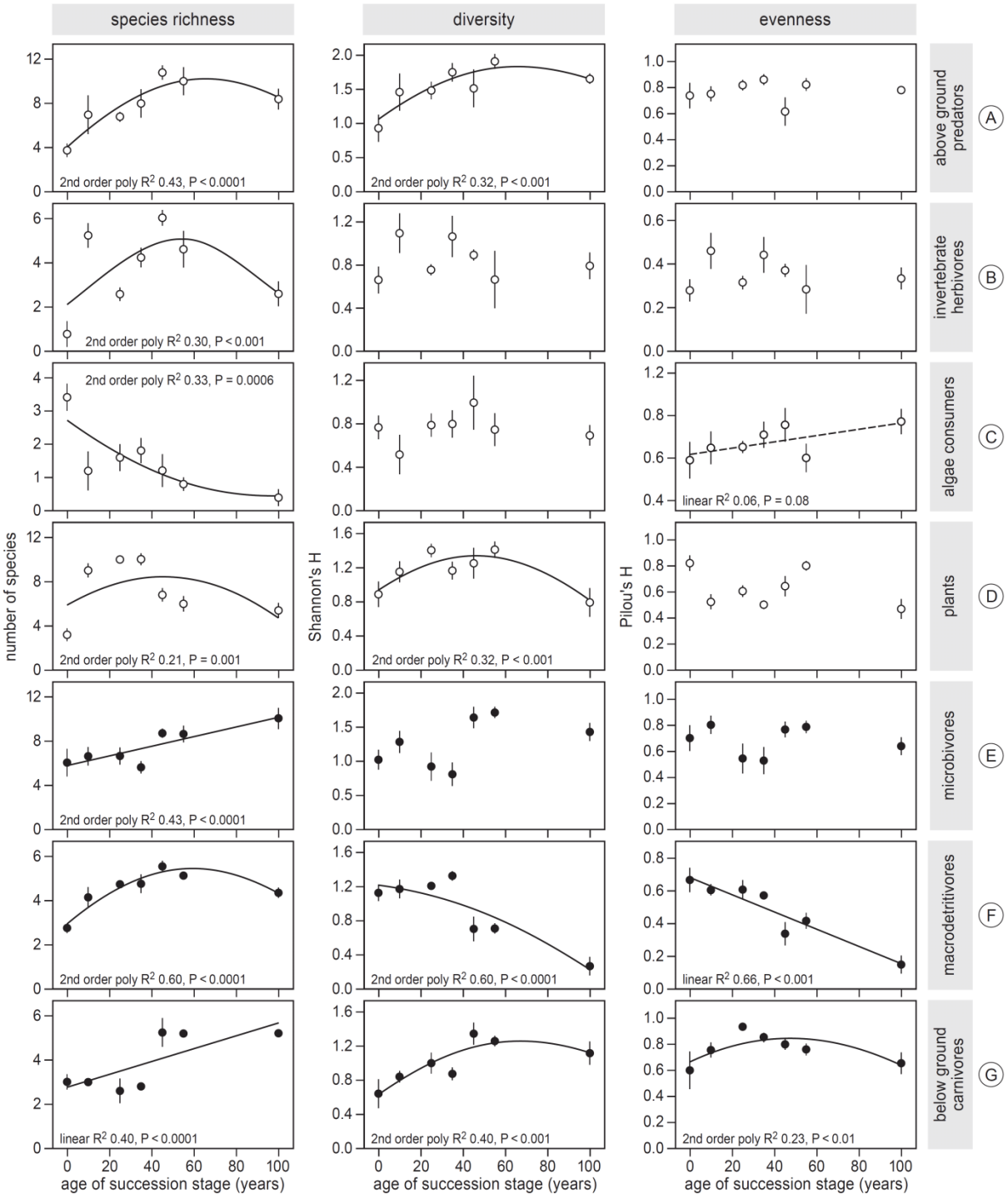

Figure 4.

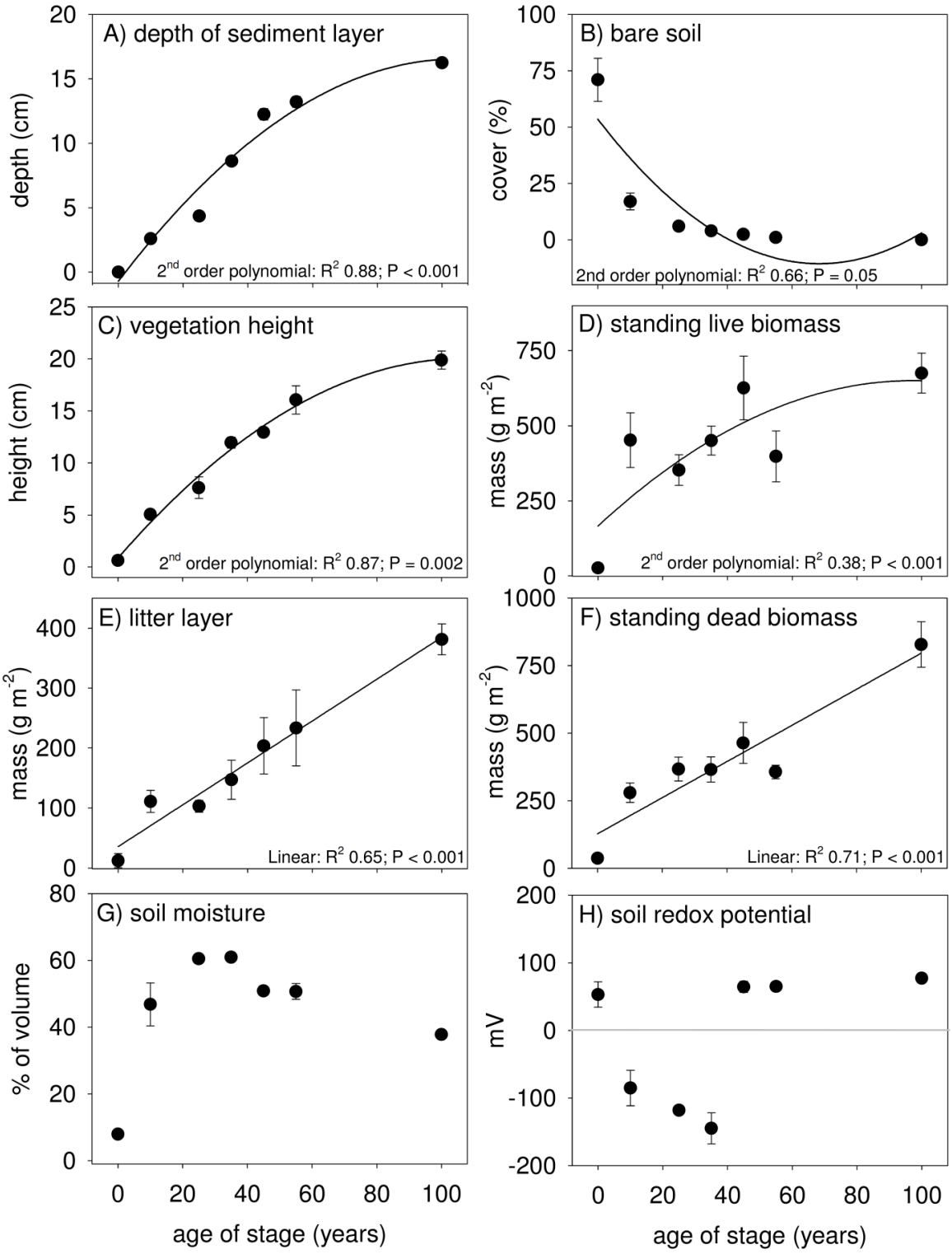

Figure 5.